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<u>L5</u>	l4 with l3	9	<u>L5</u>
<u>L4</u>	inser\$ or remov\$	5982576	<u>L4</u>
<u>L3</u>	L2 with l1	41	<u>L3</u>
<u>L2</u>	construct or vector or plasmid	570691	<u>L2</u>
<u>L1</u>	CpG with (neutralizing or immunostimula\$)	285	<u>L1</u>

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File: PGPB

May 30, 2002

PGPUB-DOCUMENT-NUMBER: 20020065236

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20020065236 A1

TITLE: CpG reduced plasmids and viral vectors

PUBLICATION-DATE: May 30, 2002

US-CL-CURRENT: [514/44](#); [435/458](#), [435/91.1](#), [435/91.4](#), [536/23.1](#)APPL-NO: 09/ 540991 [\[PALM\]](#)

DATE FILED: March 31, 2000

CONTINUED PROSECUTION APPLICATION: This is a publication of a continued prosecution application (CPA) filed under 37 CFR 1.53(d).

RELATED-US-APPL-DATA:

Application 09/540991 is a continuation-in-part-of US application 09/392462, filed September 9, 1999, PENDING

Application is a non-provisional-of-provisional application 60/099583, filed September 9, 1998,

[0001] This application is a continuation-in-part of application Ser. No. 09/392,462, filed Sep. 9, 1999, which claims priority of Provisional Application No. 60/099,583, filed Sep. 9, 1998, the disclosures of both of which are incorporated herein by reference.

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L5: Entry 3 of 9

File: PGPB

May 30, 2002

DOCUMENT-IDENTIFIER: US 20020065236 A1

TITLE: CpG reduced plasmids and viral vectors

Detail Description Paragraph:

[0052] In the present invention, a plasmid or viral vector may be modified to reduce the inflammatory response to the plasmid or vector to both reduce toxicity and increase the efficacy of gene delivery. The plasmid or vector may also be modified in order to modulate a mammal's immunostimulatory response to a plasmid or vector composition. The modified plasmid or vector may be administered alone, as the active ingredient in a formulation, or as part of a complex with a carrier such as lipids, including cationic amphiphile compounds, other viral vectors, including adenoviruses, and other methods that have been employed in the art to effectuate delivery of biologically active molecules into the cells of mammals. A plasmid:carrier complex may also be administered alone or as the active ingredient in a formulation. These elements will now be discussed. The present invention provides a method to reduce or modulate a mammal's immunostimulatory response to a composition and both reduce the toxicity and increase the efficacy of gene delivery. In the practice of the invention, a plasmid or a viral vector may be modified to reduce the inflammatory response to the plasmid or vector. In one embodiment, CpG motifs of the plasmid or vector may be methylated to reduce the immunostimulatory response. In another embodiment, CpG motifs of a plasmid or vector may be removed or altered to reduce the immunostimulatory response. In a preferred embodiment, the plasmid or vector is substantially devoid of any CpG dinucleotides.

CLAIMS:

9. A method of reducing a mammal's immunostimulatory response to a plasmid comprising the step of administering said plasmid wherein said plasmid comprises: at least one replication origin region wherein at least one CpG motif has been removed from said at least one replication origin region.

11. A method of reducing a mammal's immunostimulatory response to a viral vector comprising the step of administering said viral vector wherein at least one CpG motif is removed from said viral vector's genome.

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L5: Entry 9 of 9

File: DWPI

Nov 26, 1998

DERWENT-ACC-NO: 1999-059712

DERWENT-WEEK: 200208

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TITLE: Use of neutralising CpG and stimulating CpG motifs in DNA vectors - for enhancing the immunostimulatory effect of an antigen or enhancing the expression of a therapeutic polypeptide

Basic Abstract Text (1):

A method is claimed for enhancing the immunostimulatory effect of an antigen encoded by nucleic acid contained in a nucleic acid construct comprising: (a) determining the CpG-N and CpG-S motifs present in the construct; and (b) removing neutralising CpG (CpG-N) motifs and optionally inserting stimulatory CpG (CpG-S) motifs in the construct, thereby producing a nucleic acid construct having enhanced immunostimulatory efficacy. Also claimed are: (1) a method for stimulating a protective or therapeutic immune response to an antigen in a subject comprising administering to the subject a nucleic acid construct produced by determining the CpG-N and CpG-S motifs present in the construct and removing neutralising CpG (CpG-N) motifs and optionally inserting stimulatory CpG (CpG-S) motifs in the construct; thereby producing a nucleic acid construct having enhanced immunostimulatory efficacy and stimulating a protective or therapeutic immune response in the subject; (2) a method for enhancing the expression of a therapeutic polypeptide in vivo, where the polypeptide is encoded by a nucleic acid contained in a nucleic acid construct comprising determining the CpG-N and CpG-S motifs present in the construct, removing stimulatory CpG (CpG-S) motifs and/or inserting neutralising CpG (CpG-N) motifs, thereby producing a nucleic acid construct providing enhanced expression of the therapeutic polypeptide; (3) a method for enhancing the expression of a therapeutic polypeptide in vivo comprising administering to a subject a nucleic acid construct, where the construct is produced by determining the CpG-N and CpG-S motifs present in the construct and removing stimulatory CpG (CpG-S) motifs and/or inserting neutralising CpG (CpG-N) motifs, thereby enhancing expression of the therapeutic polypeptide in the subject.

Equivalent Abstract Text (1):

A method is claimed for enhancing the immunostimulatory effect of an antigen encoded by nucleic acid contained in a nucleic acid construct comprising: (a) determining the CpG-N and CpG-S motifs present in the construct; and (b) removing neutralising CpG (CpG-N) motifs and optionally inserting stimulatory CpG (CpG-S) motifs in the construct, thereby producing a nucleic acid construct having enhanced immunostimulatory efficacy. Also claimed are: (1) a method for stimulating a protective or therapeutic immune response to an antigen in a subject comprising administering to the subject a nucleic acid construct produced by determining the CpG-N and CpG-S motifs present in the construct and removing neutralising CpG (CpG-N) motifs and optionally inserting stimulatory CpG (CpG-S) motifs in the construct; thereby producing a nucleic acid construct having enhanced immunostimulatory efficacy and stimulating a protective or therapeutic immune response in the subject; (2) a method for enhancing the expression of a therapeutic polypeptide in vivo, where the polypeptide is encoded by a nucleic acid contained in a nucleic acid construct comprising determining the CpG-N and CpG-S motifs present in the

construct, removing stimulatory CpG (CpG-S) motifs and/or inserting neutralising CpG (CpG-N) motifs, thereby producing a nucleic acid construct providing enhanced expression of the therapeutic polypeptide; (3) a method for enhancing the expression of a therapeutic polypeptide in vivo comprising administering to a subject a nucleic acid construct, where the construct is produced by determining the CpG-N and CpG-S motifs present in the construct and removing stimulatory CpG (CpG-S) motifs and/or inserting neutralising CpG (CpG-N) motifs, thereby enhancing expression of the therapeutic polypeptide in the subject.

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L5: Entry 9 of 9

File: DWPI

Nov 26, 1998

DERWENT-ACC-NO: 1999-059712

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TITLE: Use of neutralising CpG and stimulating CpG motifs in DNA vectors - for enhancing the immunostimulatory effect of an antigen or enhancing the expression of a therapeutic polypeptide

PRIORITY-DATA: 1997US-047233P (May 20, 1997), 1997US-047209P (May 20, 1997), 1998US-0082649 (May 20, 1998)

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PATENT-FAMILY:

	PUB-NO	PUB-DATE	LANGUAGE	PAGES	MAIN-IPC
<input type="checkbox"/>	WO 9852581 A1	November 26, 1998	E	108	A61K035/00
<input type="checkbox"/>	US 6339068 B1	January 15, 2002		000	A61K048/00
<input type="checkbox"/>	AU 9876908 A	December 11, 1998		000	A61K035/00
<input type="checkbox"/>	EP 1003531 A1	May 31, 2000	E	000	A61K035/00

APPLICATION-DATA:

PUB-NO	APPL-DATE	APPL-NO	DESCRIPTOR
WO 9852581A1	May 20, 1998	1998WO-US10408	
US 6339068B1	May 20, 1997	1997US-047209P	Provisional
US 6339068B1	May 20, 1997	1997US-047233P	Provisional
US 6339068B1	May 20, 1998	1998US-0082649	
AU 9876908A	May 20, 1998	1998AU-0076908	
AU 9876908A		WO 9852581	Based on
EP 1003531A1	May 20, 1998	1998EP-0924828	
EP 1003531A1	May 20, 1998	1998WO-US10408	
EP 1003531A1		WO 9852581	Based on

INT-CL (IPC): [A61 K 35/00](#); [A61 K 48/00](#); [C12 N 15/00](#); [C12 N 15/63](#); [C12 N 15/88](#)